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## The mystery of mistletoe mitochondria

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## ABSTRACT

Respiration is one of life's most important chemical reactions as it provides energy for the cell. However the festive plant mistletoe does it differently to all other multi-celled organisms.



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Mistletoe is an evergreen parasitic plant that lives on trees and steals water and nutrients from them. It features in European folklore as a symbol for fertility and vitality, is a well-known addition to Christmas and is also known to comic fans as the main ingredient of the magic potion in Asterix the Gaul. As well as its mythical qualities it also has fascinating underlying biology.

We all breathe oxygen. This gets transported to every cell in the body, where oxygen is used to burn sugars and other nutrients, producing energy with carbon dioxide as a byproduct. This process is called respiration and is one of life's most important and central chemical reactions as it supplies energy to the rest of the cell. A key part of this process is carried out by five molecular machines, the respiratory complexes, which are housed in a compartment of the cell called a mitochondrion. These respiratory complexes are made up of lots of individual parts, for example the largest of these machines, complex I, has over 40 different proteins. All five molecular machines were thought to be essential for multi-celled life, such as animals and plants, although there are examples of single celled organisms having lost them.





That's why DNA sequences from mistletoe were puzzling, as it suggested this plant species may have lost complex I. The majority of the cell's DNA resides in the nucleus. Two studies, one from a group in the USA and another in Denmark, looked at the genes inside the mitochondria, which house a small number of genes. They found that the genes needed to make complex I were missing, strongly implying it had been lost. If this were true it would be the first multi-celled organism to have lost complex I. However, it remained possible that the genes had been transferred to the nucleus. The genome of mistletoe has not been sequenced because it is very large and of no commercial interest. Therefore, we decided to collect mitochondria from mistletoe and look inside to see if complex I really had been lost and, if so, how the mitochondria had been adapted to cope with this upheaval.

We collected branches of mistletoe from trees in both the U.K. and Germany and took them back to the lab. Through a combination of blending leaf buds using a kitchen blender and spinning the sample at different speeds, we separated out the different parts of the cell and isolated mitochondria. Now we could test what is inside. As a basis for comparison we used another plant that scientists understand very well called *Arabidopsis* (Thale Cress). We have numerous biochemical techniques for studying the respiratory complexes, for example, we can use specific staining methods to determine their abundance. Additionally, we can get a list of all the different proteins in a sample using a technique called mass spectrometry.

Using these techniques we found no trace of complex I. In addition, we also observed that the

other respiratory complexes were of much lower abundance compared to Arabidopsis. This would suggest that the amount of energy generated in the mitochondria is very low in mistletoe. This raises the question of how the mistletoe copes with such low energy production from the mitochondria? We measured how much carbon dioxide is released by different metabolic reactions and found a much greater rate of an alternative energy pathway, called glycolysis. Glycolysis generates energy in a different part of the cell. It is less efficient than respiration but can still produce enough energy for the cell if its substrates, simple sugars, are abundant. This implies that mistletoe has remodeled its energy metabolism and that, as a parasitic plant, it uses the sugars stolen from its host to cope with the loss of a key piece of molecular machinery.

Mistletoe is the first multi-celled organism to have lost complex I, forcing us to rethink how important the respiratory complexes are. Is mistletoe a unique case, or are there other parasitic plants out there that respire differently? The main outstanding question is why did complex I get lost in the first place? It could be connected to the mistletoe's parasitic lifestyle. It is commonly seen that parasites can lose seemingly key pieces of the metabolism, as they can get what they need from their host. For now, the mitochondria of mistletoe remain mysterious.

The lack of complex I in mistletoe mitochondria was also shown by the group of Hans-Peter Braun. The publications appeared together in the same issue of Current Biology.